AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the application:

1. (original) A process for the quantitative determination of 25-hydroxy-cholecalciferol in animal feed which comprises the steps of

a) dispersing the feed sample in water and adding to the sample a defined amount of an internal standard compound having a mass different from 25-hydroxycholecalciferol and having a polarity similar to but different from 25-hydroxycholecalciferol;

b) extracting the aqueous dispersion with tert. butyl methyl ether;

c) submitting the ether extract to semipreparative HPLC;

d) collecting the fractions containing 25-hydroxycholecalciferol and the internal standard compound;

e) submitting the fractions collected in d) or an aliquot thereof to HPLC combined with mass spectrometry;

f) determining the MS peak areas of 25-hydroxycholecalciferol and of the internal standard compound added; and

g) calculating the amount of 25-hydroxycholecalciferol by computing the MS peak areas measured.

2. (original) A process as in claim 1 wherein the standard compound is 26,27-hexadeutero-25-hydroxycholecalciferol, 25-hydroxy-ergocalciferol, or 1α-hydroxy-cholecalciferol.

3. (original) A process as in claim 2 wherein the standard compound is 26,27-hexadeutero-25-hydroxycholecalciferol.

4. (currently amended) A process as in ~~any one of claims 1-3~~ claim 1 wherein the semipreparative HPLC is carried out on silica gel as the stationary phase and an isopropanol:ethyl acetate:isooctan mixture as the mobile phase.

5. (original) A process as in claim 4 wherein the mobile phase is isopropanol:ethyl acetate:isooctan in a ratio (by volume) of about 1 : 10 : 89.

6. (currently amended) A process as in claim 4 ~~or 5~~ wherein the stationary phase is Hypersil Si 60, 3 µm.

7. (currently amended) A process as in ~~any one of claims 1-6~~ claim 1 wherein the analytical HPLC is carried out in a chromatography system comprising a trapping column on which the substances to be measured are concentrated, and the intrinsic analytical column for separation.

8. (original) A process as in claim 4 wherein the stationary phase in the analytical HPLC is a modified silica gel such as Aquasil C18, 3 µm.

9. (currently amended) A process as in claim 7 ~~or 8~~ wherein a gradient of water containing 0.05 % (vol/vol) formic acid and methanol containing 0.05 % (vol/vol) formic acid is used as the mobile phase.